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TITEL: METHOD OF ANALYSING GRANULAR COMPOSITION BY ACOUSTIC EMISSION

FIELD OF THE INVENTION

The present invention relates to a method for analyzing a property of a granular composition comprising an active compound by subjecting the granular composition to acoustic emission analysis. The invention also relates to a method for producing a particulate product comprising subjecting the product to acoustic emission analysis. Further the invention relates to a granulation and/or coating apparatus suitable for preparing a granular composition comprising an active compound said apparatus comprising means for performing acoustic emission analysis.

BACKGROUND OF THE INVENTION

Acoustic emission methods are, *inter alia*, known from Whitaker et al. *Application of acoustic emission to the monitoring and end point determination of a high shear granulation process*, Int. J. Pharm., 205, pp 79-91, 2000. Methods of using acoustic emission analysis e.g. on sand powders are known from Esbensen K et al, Acoustic chemometrics - from noise to information, Chemometrics and intelligent laboratory systems, 44 (1998) 61-76. Methods of using high frequency acoustic emission analysis in fluid beds are known from Tsujimoto H et al, Monitoring particle fluidization in a fluidized bed granulator with an acoustic emission sensor, Powder technology, 113 (2000) 88-96.

SUMMARY OF THE INVENTION

The present invention relates to a method for analyzing a property of a granular composition/granules comprising a

biologically active compound by subjecting the granular composition to acoustic emission analysis. Formulation of chemical compounds into finished goods, in particular granulation, is usually required to achieve improved properties of the products, thus making them more commercially attractive. However, for biologically active compounds, granulation is often compulsory to the producers because the active compound must, until being applied in the intentional use, be separated from the surrounding environment to ensure the safe handling of the product. The amount of biologically active compound which can escape from the granulated product, e.g. in the form of dust, must be minimized to ensure that persons handling the product do not suffer any adverse effects from contact with the biologically active compound. Vice versa the active compound must be protected from the environment outside the granule to remain stabile and active once it is to be used. Once an active compound has been granulated it is known that one may further coat granules comprising biologically active compound with a coating agent which further suppress the release of active compound from the granule and further improve the stability of the active compound in the granule.

One object of the invention is, to provide methods for detecting, in a granular composition comprising a biologically active compound, the amount of active compound released from granules in the form of active dust, during or after the process for preparing the granular composition. Another object of the invention is to design a granulation apparatus and to select method setup, so that the method may be used on-line or in-line in the production of such granular compositions, and that the methods of the invention may in real time may provide information about levels of dust comprising biologically active compound during processing of the granular composition. A further object of the invention is to provide, during or after a

process of coating of granules comprising a biologically active compound, methods for monitoring the thickness and/or integrity of coating layers applied to the granules to suppress dust formation and increase the stability of the active compound.

We have found that in the process control of the making of granular compositions comprising a biologically active compound not only the quality of granules formed may by evaluated also the amount of small dust particles present in the granular composition e.g. as a result from release of dust from the granules or as a result from insufficient granulation can be evaluated by recording 20Hz-20 kHz vibration signals arising from granules and dust of the granular composition colliding with the granulator steel walls and subjecting the recorded vibration signals to computerized data processing.

We have also found that in the process of making coated granular compositions comprising a biologically active compound the thickness and/or homogeneity of a coating deposited on the granules of the granular composition e.g. as a result from applying a coating agent to the granules can be evaluated by recording 20Hz to 20kHz sound signals arising from coated granules colliding with the granulator steel walls and subjecting the recorded vibration signals to computerized data processing.

Accordingly, the present invention provides in a first aspect a method for acoustic emission analysis of a granular composition comprising a biologically active compound, said method comprising colliding the granular composition with at least one surface transmitting low frequency vibrations, recording low frequency vibration data in range of 20 Hz to 20 kHz arising from the collision with at least one vibration detector and subjecting the recorded low frequency vibration data to computerized data processing.

Further, in a second aspect, the invention provides a process for preparing granules comprising a biologically active compound and optionally auxiliary granulation agents in a granulation apparatus said process comprising the step of performing acoustic emission analysis in accordance with the first aspect of the invention (*vide supra*).

Still further, in a third aspect, the invention provides a granulation or coating apparatus comprising (a) a granulating or coating device comprising at least one chamber for granulating material or for coating of granulated material and at least surface capable of transmitting low frequency sound, (b) at least one detector capable of detecting low frequency vibration, (c) means recording low frequency vibration signals detected by the detector, (d) means processing recording the low frequency vibration signals.

Finally, in a fourth aspect the invention provides use of acoustic emission analysis on granules comprising a biologically active compound.

BRIEF DESCRIPTION OF THE TABLES AND DRAWING

Figure 1: Score plot obtained when changing the supply of filler to a continuous high shear mixer granulation process

DETAILED DESCRIPTION OF THE INVENTION

Acoustic emission analysis on granules

The present invention relates, as described, to a method for performing acoustic emission analysis on a granular composition comprising an active compound. As a particle collides with a hard surface the particle will interfere with the surface and make the surface vibrate. The basic idea is that these vibrations give information of the characteristics

of the particles. Accordingly, if the characteristics of the particles change the vibrations will also change.

The first step of the method comprises colliding the granular composition to be analyzed with a hard surface. In a granulation process the granules in a granulation apparatus will collide with walls of the granulation chamber as long as movement is induced to granules.

The second step of the method comprises the recording of vibration caused by the collision between granules and the walls of the granulation apparatus. In a preferred embodiment the one or more accellerometers capable of detecting vibrations in the range of 20 Hz to 20 kHz, preferably 100 Hz to 10 kHz, more preferably 500 Hz to 5 kHz is positioned directly on the wall of the granulation apparatus. The accelerometer is connected to a computer unit capable of storing the recorded vibration data

For analyzing the stored vibration data, these data are firstly subjected to Fast Fourier Transformation to reduce noise. Then the transformed data are analyzed by Principal Component Analysis (PCA) using a standard software for that purpose, such as Unscrambler® from Camo, Norway. In this operation the data is arranged into score plots. From the score plot it can be revealed if a change in the granular composition has occurred in the granulation process, such as change in properties of the granules, changes in coatings or formation of dust.

The granular composition

Physical properties

The granular composition of the invention is a composition comprising the biologically active compound and optionally auxiliary granulation agents and coating agents processed into

particles or granules. Accordingly, finished granules are the result of the processes and methods of the invention. The term "granules" are to be understood as a predominantly spherical or near spherical structure of a macromolecular size, preferably having an average size measure in the longest diameter between 20-2000 μm, more preferably between 100-1000 μm, most preferably between 200-800 μm. The spherical granules preferably have a ratio, (a):(b), between the diameter in the shortest dimension (a) and the diameter in the longest dimension (b) of the granule of between 1:1 to 1:5, preferably between 1:1 to 1:3.

The "size distribution" (PSD) of granules can be expressed in terms of the mass mean diameter of the individual particles. A mean mass diameter of D50 is the diameter at which 50% of the granules, by mass, have a smaller diameter, while 50% by mass have a larger diameter. The values D10 and D90 are the diameters at which 10% and 90%, respectively, of the granules, by mass, have a smaller diameter than the value in question. The "SPAN" indicates the breadth of the PSD and is expressed as:
(D90-D10)/D50. For purposes of the present invention, the PSD of granules after granulation is normally as narrow as possible. Use of acoustic emission analysis, in accordance with the present invention, for controlling the granulation process may aid in lowering of the PSD, and the SPAN of the granular composition after granulation is therefore preferably less than about 2.5, preferably less than about 2.0, more preferably less than about 1.5, and most preferably less than about 1.0.

The granules are preferably coated with a coating agent forming a, preferably homogenous, coherent and continuous, layer around the granules. The term coating agent as used herein is to be understood as single coating compound or a

mixture of coating compounds. Coated granules thus consist of a granule core and a granule coating. Preferably the coating layer is relatively thick in order to further reduce dusting and improve stability of the biologically active compound. The coating thickness may be described by the ratio between the average diameter of a coated granule core and the average diameter of an uncoated granule core (hereinafter abbreviated D_G/D_C), i.e. the average diameter of the coated granule divided by the average diameter of the granule core only. If for example a granule core having a diameter of 100 μm is coated with a coating layer 200 μm thick, the granule would have a diameter of (200+100+200)=500 μm and D_G/D_C is 500 μm/100 μm = 5. Coated granules of the invention preferably have a D_G/D_C of at least 1.1, which means that the thickness of the coating is at least 5% of the average granule core diameter. A more preferred D_G/D_C is at least 1.5, more preferably at least 2, more preferably at least 2.5, more preferably at least 3, most preferably at least 4. D_G/D_C is however preferably below about 100, preferably below about 50, more preferably below 25, and most preferably below 10. A most preferred range for D_G/D_C is about 4 to about 6.

Furthermore, in the present invention the coating is substantially free of biologically active compound. The term "substantially free of biologically active compound" as used herein about a coating means that there less than 5 mg of biologically active compound per gram coating agent.

Construction

The construction of the granules of the invention may be divided into the following non-exhaustive categories:

a) Spray dried granules, wherein a liquid solution containing the biologically active compound is atomized in a spray dryer o form small droplets which during their way down the dryer

dry to form a granular material comprising the active compound. Very small granules can be produced this way (Michael S. Showell (editor); *Powdered detergents*; Surfactant Science Series; 1998; vol. 71; page 140-142; Marcel Dekker). For these granules the active compound is intimately mixed with any other auxiliary granulation agents present in the liquid solution.

b) Layered granules, wherein the biologically active compound is coated as a layer around a pre-formed core particle, wherein an solution containing the biologically active compound, and preferably auxiliary granulation agents, is atomized, typically in a fluid bed apparatus wherein the pre-formed core particles are fluidized, and the solution of active compound adheres to the core particles and dries up to leave a layer of dry biologically active compound on the surface of the core particle. Granules of a desired size can be obtained this way if a useful core particle of the desired size can be found. This type of granules is described in e.g. WO 97/23606

c) Absorbed core granules, wherein rather than coating the biologically active compound as a layer around the core, the biologically active compound is absorbed onto and/or into the surface of the core. Such a process is described in WO 97/39116.

d) Extruded or pelletized granules, wherein a paste containing the biologically active compound is pressed into granules in a mould or under pressure is extruded through a small opening and cut into granules which are subsequently dried. Such granules usually have a considerable size because of the material in which the extrusion opening is made (usually a

plate with bore holes) sets a limit on the allowable pressure drop over the extrusion opening. Also, very high extrusion pressures when using a small opening increase heat generation in the paste, which may be harmful to the biologically active compound. (Michael S. Showell (editor); *Powdered detergents*; Surfactant Science Series; 1998; vol. 71; page 140-142; Marcel Dekker)

e) Spray cooled granules, wherein a powder of biologically active compound is suspended in molten wax and the suspension is sprayed, e.g. through a rotating disk atomizer, into a cooling chamber where the droplets quickly solidify (Michael S. Showell (editor); *Powdered detergents*; Surfactant Science Series; 1998; vol. 71; page 140-142; Marcel Dekker). For these granules the active compound is intimately mixed with the wax instead of being concentrated on its surface. Also US 4,016,040 and US 4,713,245 are documents relating to this technique

f) High shear mixer granules, wherein a liquid containing the biologically active compound is added to a dry powder composition of auxiliary granulation agent. The liquid and the powder in a suitable proportion are mixed and as the moisture of the liquid is absorbed in the dry powder, the components of the dry powder will start to adhere and agglomerate and granules will build up, forming granules comprising the biologically active compound. For these granules the active compound is intimately mixed with the auxiliary granulation agents. Such a process is described in US 4,106,991 (NOVO NORDISK) and related documents EP 170360 B1 (NOVO NORDISK), EP 304332 B1 (NOVO NORDISK), EP 304331 (NOVO NORDISK), WO 90/09440 (NOVO NORDISK) and WO 90/09428 (NOVO NORDISK).

Dust particles in granular compositions

Dust particles, which may be present in a granular composition, may be characterized in that they are particles, which usually have a considerably smaller size than the granules and do not possess the characteristic spherical shape of the granules. Dust particles typically have an irregular non-spherical and abrupt structure such as rod or flake shaped. Dust particles are typically much smaller than the average size of granules, and most dust particles are, depending on the granular composition less than 20 μm in diameter. Accordingly, without being bound to the theory it is presently contemplated that these physical difference between granules and dust particles is contributing to the differences in low frequency sound arising when colliding the granules and dust particles with a surface.

Compounds in the granular composition

Biologically active compounds

The granular composition of the invention comprises a biologically active compound, preferably in a purified form. The term biologically active compound as used herein is to be understood as any compound, which is active in a biological system such as compounds, which interfere with and/or modifies biological pathways or biological reactions. The term "purified" as used herein is to be understood as biologically active compounds, which before granulation has been subjected to one or more purification step to remove e.g. excess material and/or to concentrate the active compound. In the case the active compound is prepared by a microbiological fermentation process purification preferably includes step selected from filtering, ultra-filtration, flocculation, sedimentation, evaporation, extraction and the like, to remove biomass and other undesired

matter including water to yield a mixture which is enriched in the biologically active compound.

Biologically active compounds include among others organic compounds such as bio-catalysts, therapeutic agents, herbicides, pesticides and fungicides. Preferred biologically active compounds are producible by fermenting a microorganism producing the active compounds.

Preferred compounds are those among proteins and peptides, more preferably catalytic proteins, i.e. enzymes, because proteins such as enzymes are used in vast volumes in industry and are known to cause adverse allergy reactions in humans or animal when exposed to such proteins. Furthermore, enzymes are widely used in household products such as detergents for removing soil of a biological origin, and many industrial processes involves human handling of the enzymes. The enzyme may be any enzyme for which it is desired to separate the enzyme from the surrounding environment through granulation of the enzymes.

The enzyme classification employed in the present specification with claims is in accordance with *Recommendations (1992) of the Nomenclature Committee of the International Union of Biochemistry and Molecular Biology*, Academic Press, Inc., 1992.

Accordingly the types of enzymes which may appropriately be incorporated in granules of the invention include oxidoreductases (EC 1.-.-.-), transferases (EC 2.-.-.-), hydrolases (EC 3.-.-.-), lyases (EC 4.-.-.-), isomerases (EC 5.-.-.-) and ligases (EC 6.-.-.-).

Preferred oxidoreductases in the context of the invention are peroxidases (EC 1.11.1), laccases (EC 1.10.3.2) and glucose oxidases (EC 1.1.3.4)], while preferred transferases are transferases in any of the following sub-classes:

a) Transferases transferring one-carbon groups (EC 2.1);
b) Transferases transferring aldehyde or ketone residues (EC 2.2); acyltransferases (EC 2.3);
c) Glycosyltransferases (EC 2.4);
d) Transferases transferring alkyl or aryl groups, other that methyl groups (EC 2.5); and
e) Transferases transferring nitrogenous groups (EC 2.6).

A most preferred type of transferase in the context of the invention is a transglutaminase (protein-glutamine γ-glutamyltransferase; EC 2.3.2.13).

Further examples of suitable transglutaminases are described in WO 96/06931 (Novo Nordisk A/S).

Preferred hydrolases in the context of the invention are: Carboxylic ester hydrolases (EC 3.1.1.-) such as lipases (EC 3.1.1.3); phytases (EC 3.1.3.-), e.g. 3-phytases (EC 3.1.3.8) and 6-phytases (EC 3.1.3.26); glycosidases (EC 3.2, which fall within a group denoted herein as "carbohydrases"), such as α-amylases (EC 3.2.1.1); peptidases (EC 3.4, also known as proteases); and other carbonyl hydrolases].

In the present context, the term "carbohydrase" is used to denote not only enzymes capable of breaking down carbohydrate chains (e.g. starches) of especially five- and six-membered ring structures (i.e. glycosidases, EC 3.2), but also enzymes capable of isomerizing carbohydrates, e.g. six-membered ring structures such as D-glucose to five-membered ring structures such as D-fructose.

Carbohydrases of relevance include the following (EC numbers in parentheses):

α-amylases (3.2.1.1), β-amylases (3.2.1.2), glucan 1,4-α-glucosidases (3.2.1.3), cellulases (3.2.1.4), endo-1,3(4)-β-glucanases (3.2.1.6), endo-1,4-β-xylanases (3.2.1.8), dextranases (3.2.1.11), chitinases (3.2.1.14), polygalacturonases (3.2.1.15), lysozymes (3.2.1.17), β-

glucosidases (3.2.1.21), α-galactosidases (3.2.1.22), β-galactosidases (3.2.1.23), amylo-1,6-glucosidases (3.2.1.33), xylan 1,4-β-xylosidases (3.2.1.37), glucan endo-1,3-β-D-glucosidases (3.2.1.39), α-dextrin endo-1,6-α-glucosidases (3.2.1.41), sucrose α-glucosidases (3.2.1.48), glucan endo-1,3-α-glucosidases (3.2.1.59), glucan 1,4-β-glucosidases (3.2.1.74), glucan endo-1,6-β-glucosidases (3.2.1.75), arabinan endo-1,5-α-L-arabinosidases (3.2.1.99), lactases (3.2.1.108), chitosanases (3.2.1.132) and xylose isomerases (5.3.1.5).

Examples of commercially available oxidoreductases (EC 1.-.-.-) include Gluzyme™ (enzyme available from Novo Nordisk A/S). Examples of commercially available proteases (peptidases) include Kannase™, Everlase™, Esperase™, Alcalase™, Neutrase™, Durazym™, Savinase™, Pyrase™, Pancreatic Trypsin NOVO (PTN), Bio-Feed™ Pro and Clear-Lens™ Pro (all available from Novo Nordisk A/S, Bagsvaerd, Denmark).

Other commercially available proteases include Maxatase™, Maxacal™, Maxapem™, Opticlean™ and Purafect™ (available from Genencor International Inc. or Gist-Brocades).

Examples of commercially available lipases include Lipoprime™ Lipolase™, Lipolase™ Ultra, Lipozyme™, Palatase™, Novozym™ 435 and Lecitase™ (all available from Novo Nordisk A/S).

Other commercially available lipases include Lumafast™ (*Pseudomonas mendocina* lipase from Genencor International Inc.); Lipomax™ (*Ps. pseudoalcaligenes* lipase from Gist-Brocades/Genencor Int. Inc.; and *Bacillus* sp. lipase from Solvay enzymes.

Examples of commercially available carbohydrases include Alpha-Gal™, Bio-Feed™ Alpha, Bio-Feed™ Beta, Bio-Feed™ Plus, Bio-Feed™ Plus, Novozyme™ 188, Celluclast™, Cellusoft™,

Ceremyl™, Citrozym™, Denimax™, Dezyme™, Dextrozyme™, Finizym™, Fungamyl™, Gamanase™, Glucanex™, Lactozym™, Maltogenase™, Pentopan™, Pectinex™, Promozyme™, Pulpzyme™, Novamyl™, Termamyl™, AMG™ (Amyloglucosidase Novo), Maltogenase™, Sweetzyme™ and Aquazym™ (all available from Novo Nordisk A/S). Further carbohydrases are available from other suppliers.

The amount of enzyme to be incorporated in a granule of the invention will depend on the intended use of the granule. For many applications, the enzyme content will be as high as possible or practicable.

The content of enzyme (calculated as pure enzyme protein) in a granule of the invention will typically be in the range of from about 0.5% to 50% by weight of the enzyme-containing granule.

Auxiliary granulation agents

The granules of the invention preferably contains auxiliary granulation agents for purposes such as aiding the formation of granules, controlling density and volume of granules, controlling amount of active compound in the granules, stabilizing the active compound and the like.

Auxiliary granulating agents may include but is not limited to:

a) Fillers such as fillers conventionally used in the field of granulation e.g. water soluble and/or insoluble inorganic salts such as finely ground alkali sulphate, alkali or earth alkali carbonate and/or alkali chloride), clays such as kaolin (e.g. Speswhite™, English China Clay), bentonites, talcs, zeolites, and/or silicates.

b) Binders such as binders conventionally used in the field of granulation e.g. binders with a high melting point or no melting

point at all and of a non waxy nature e.g. polyvinyl pyrrolidon, dextrins, polyvinylalkohol, cellulose derivatives, for example hydroxypropyl cellulose, methyl cellulose or CMC. A suitable binder is a carbohydrate binder such as Glucidex 21D available from Roquette Freres, France.

c) Fiber materials such as fibers conventionally used in the field of granulation. Pure or impure cellulose in fibrous form can be sawdust, pure fibrous cellulose, cotton, or other forms of pure or impure fibrous cellulose. Also, filter aids based on fibrous cellulose can be used. Several brands of cellulose in fibrous form are on the market, e.g. CEPO and ARBOCELL. In a publication from Svenska Trämjolsfabrikerna AB, "Cepo Cellulose Powder" it is stated that for Cepo S/20 cellulose the approximate maximum fiber length is 500 μm, the approximate average fiber length is 160 μm, the approximate maximum fiber width is 50 μm and the approximate average fiber width is 30 μm. Also, it is stated that CEPO SS/200 cellulose has an approximate maximum fiber length of 150 μm, an approximate average fiber length of 50 μm, an approximate maximum fiber width of 45 μm and an approximate average fiber width of 25 μm. Cellulose fibers with these dimensions are very well suited for the purpose of the invention. The words "Cepo" and "Arbocel" are Trademarks. Preferred fibrous cellulose is Arbocel™ BFC200. Also synthetic fibres may be used as described in EP 304331 B1 and typical fibres may be made of polyethylene, polypropylene, polyester, especially nylon, polyvinylformat, poly(meth)acrylic compounds.

d) Liquid agents such as conventionally used in the field of granulation. A liquid agent is used in conventional mixer granulation processes for enabling the build up or agglomeration of the conventional granulating component particles into

granules. The liquid agent is water and/or a waxy substance. The liquid agent is always used in a liquid phase in the granulation process but may later on solidify; the waxy substance if present, therefore, is either dissolved or dispersed in the water or melted. By the term "waxy substance" as used herein is meant a substance which possesses all of the following characteristics 1) the melting point is between 30 and 100°C, preferably between 40 and 60°C, 2) the substance is of a tough and not brittle nature, and 3) the substance possesses a certain plasticity at room temperature. Both water and waxy substance are liquid agents, i.e. they are both active during the formation of the granules; the waxy substance stays as a constituent in the finished granules, whereas the majority of the water is removed during a drying step. Examples of waxy substances are polyglycols, fatty alcohols, ethoxylated fatty alcohols, mono-, di- and triglycerolesters of higher fatty, acids, e.g. glycerol monostearate, alkylarylethoxylates, and coconut monoethanolamide.

If a high amount of waxy substance is used, relatively little water should be added, and vice versa. Thus, the liquid agent can be either water alone, waxy substance alone or a mixture of water and waxy substance. When a mixture of water and waxy substance is used the water and the waxy substance can be added in any sequence, e.g. first the water and then the waxy substance, or first the waxy substance and then the water or a solution or suspension of the waxy substance in the water. Also, when a mixture of water and waxy substance is used, the waxy substance can be soluble or insoluble (but dispersible) in water. If water is used a liquid agent it may not be a part of the finished mixer granule as usually most of the water is dried off at a subsequent drying of the mixer granules.

e) Enzyme stabilizing or protective agents such as conventionally used in the field of granulation. Stabilizing or protective agents may fall into several categories: alkaline or neutral materials, reducing agents, antioxidants and/or salts of first transition series metal ions. Each of these may be used in conjunction with other protective agents of the same or different categories. Examples of alkaline protective agents are alkali metal silicates, -carbonates or bicarbonates, which provide a chemical scavenging effect by actively neutralizing e.g. oxidants. Examples of reducing protective agents are salts of sulfite, thiosulfite or thiosulfate, while examples of antioxidants are methionine, butylated hydroxytoluene (BHT) or butylated hydroxyanisol (BHA). Most preferred agents are salts of thiosulfates, e.g. sodium thiosulfate. Also enzyme stabilizers may be borates, borax, formates, di- and tricarboxylic acids and reversible enzyme inhibitors such as organic compounds with sulfhydryl groups or alkylated or arylated boric acids.

f) Cross-linking agents such as conventionally used in the field of granulation. Cross-linking agents may be enzyme-compatible surfactants e.g. ethoxylated alcohols, especially ones with 10 to 80 ethoxy groups.

Further, suspension agents, mediators (for boosting bleach action upon dissolution of the granule in e.g. a washing application or mediators for enzymes) and/or solvents may be incorporated as auxiliary granulating agents.

Coating agents

The coating comprises one or more conventional coating agents components such as described in WO 89/08694, WO 89/08695, EP 270 608 B1 and/or WO 00/01793. Other examples of coating agents may be found in US 4,106,991, EP 170360, EP 304332, EP 304331, EP 458849, EP 458845, WO 97/39116, WO 92/12645A, WO

89/08695, WO 89/08694, WO 87/07292, WO 91/06638, WO 92/13030, WO 93/07260, WO 93/07263, WO 96/38527, WO 96/16151, WO 97/23606, US 5,324,649, US 4,689,297, EP 206417, EP 193829, DE 4344215, DE 4322229 A, DD 263790, JP 61162185 A and/or JP 58179492. Especially the salt coatings described in WO 00/01793 are useful for coatings in the present invention.

The coating agent may be selected from the list of auxiliary granulation agents described, *supra*. Further coating agents may be selected the following non-limiting list of polymers, chlorine scavengers, plasticizers, pigments, lubricants (such as surfactants or antistatic agents) and fragrances.

Polymers useful in coating layers include vinyl polymers or vinyl co-polymers such as polyvinyl alcohol (PVA) and/or polyvinyl pyrrolidone or derivatives thereof. Also included are isoptalic acid polymers.

Plasticizers useful in coating layers in the context of the present invention include, for example: polyols such as sugars, sugar alcohols, or polyethylene glycols (PEGs) having a molecular weight less than 1000; urea, phthalate esters such as dibutyl or dimethyl phthalate; and water.

Suitable pigments include, but are not limited to, finely divided whiteners, such as titanium dioxide or kaolin, coloured pigments, water soluble colorants, as well as combinations of one or more pigments and water soluble colorants.

As used in the present context, the term "lubricant" refers to any agent, which reduces surface friction, lubricates the surface of the granule, decreases tendency to build-up of static electricity, and/or reduces friability of the granules. Lubricants can also play a related role in improving the coating process, by reducing the tackiness of binders in the coating. Thus, lubricants can serve as anti-

agglomeration agents and wetting agents. Examples of suitable lubricants are polyethylene glycols (PEGs) and ethoxylated fatty alcohols.

In embodiments aimed primarily at granules for detergent formulations, different "functional" components could be added to the coating such as TAED, CMC, bleach, OBA, surfactants, perfume as well as other functional components used in detergent formulations known to the person skilled in the art. The coating may also optionally comprise functional components selected for their specific use in the, pharmaceutical industry, agriculture, foodstuffs industry, baking industry, additives industry, feed industry, detergents industry or other industries where granules comprising a biologically active compound can be used.

In a preferred embodiment of the invention the granule of the invention is coated with a protective coating having a high constant humidity of at least 81% such as described in WO 89/08694, which is hereby incorporated by reference. Accordingly, the coating should, in certain embodiments, act as a moisture and/or bleach barrier to stabilize the biologically active compound in the core. Furthermore, in alternative embodiments, the coating unit acts as a mechanical barrier during mechanical processes such as dosing or tabletting. In certain embodiments, the coating unit is sufficiently compressible and flexible for the core to withstand a tabletting process, both in a structural sense and with regards to biological activity of the active compound. This is potentially most applicable for detergent formulations.

Acoustic emission analysis in granulation and coating processes

The present invention also encompass processes for preparing granular compositions comprising a biologically active compound and optionally auxiliary granulation agents in a granulation apparatus using the above mentioned method of acoustic emission analysis to predict properties of the granular composition and control and improve the preparation process.

Accordingly, the present invention provides a process for preparing granules comprising a biologically active compound and optionally auxiliary granulation agents in a granulation apparatus said process comprising the step of performing acoustic emission analysis on the granular composition as described, *supra*, on the granules forming in the granulation apparatus.

In a preferred embodiment the acoustic emission analysis is carried out during the formation of granules in the granulation process, preferably on-line, meaning that the acoustic emission analysis is performed more than one time in real time during the granulation process with a suitable rate of repetition. The repetition rate will, *inter alia*, depend on the data processing of data from the detector(s). In the preferred embodiment of using an accellerometer about several recordings per second is recorded. The term "formation of granules" includes also coating granules with a coating layer. In this embodiment the process also preferably comprises the step of changing at least one process parameter as a result of the acoustic emission analysis. The process parameter to be changed may be any parameter influencing the granulation process and/or the properties of the formed granules. These parameters may be supply of granulation material, i.e. biologically active compound and/or auxiliary granulation

agents and/or coating agent to the granulator, supply of gas to the granulator, temperature in the granulator, pressure in the granulator, pH in the granulator and mechanical force conferred to the granulation material. The process parameter may be changed manually or though an automated system, cf. granulation apparatus.

In a further additional embodiment acoustic emission analysis in accordance with the invention may also suitably be used to control dusting properties of finished granular compositions after granulation. Accordingly, the invention further provides a method for acoustic emission analysis of dust in a granular composition comprising a biologically active compound. Using this method, granular compositions, which do not meet the desired quality with respect to dust, may be discarded or reprocessed.

In a further additional embodiment acoustic emission analysis in accordance with the invention may also suitably be used to control coating thickness and/or homogeneity of finished granular compositions after granulation. Accordingly, the invention further provides a method for acoustic emission analysis of coating thickness in a composition of coated granules comprising a biologically active compound. Using this method granular compositions, which do not meet the desired quality with respect to coating thickness be discarded or reprocessed.

Granulation apparatus

Also included in the scope of the invention is a granulation and/or coating apparatus comprising means for performing acoustic emission analysis on granular compositions in accordance with the invention. Accordingly, the invention provides a granulation or coating apparatus comprising:

(a) a granulation or coating device comprising at least one chamber for processing material into granules or coated

granules and at least one surface transmitting low frequency sound

(b) an arrangement for performing acoustic emission analysis comprising at least one vibration detector capable of detecting vibrations in the range of 20 Hz-20 kHz and optionally amplifying and filtering units and a computer unit.

The granulation or coating device may be any conventional granulation device is preferably selected from fluid bed granulators or coaters, high shear mixer granulators, coating mixers, spray dryers, a spray coolers and extruders.

As indicated above the arrangement for performing acoustic emission analysis is suitably connected to the granulation or coating device to enable on-line or at-line acoustic emission analysis of granular compositions. On-line analysis is to be understood as analysis performed on granules as they are actually being granulated, e.g. by analyzing granules in the granulator or in a recycled purge stream. At-line analysis is to be understood as analysis performed down stream after the granulation process (e.g. at the outlet) or on non-recycled samples taken from the granulator during granulation.

The granulation apparatus may comprise other elements such computing units for processing data from detectors, optionally equipped with specialized data handling hardware and software. The granulation apparatus may also comprise control units linked to the computing units for controlling and adjusting the granulation process based on the results of the acoustic emission analysis. A control unit may be a PC, PLC or other equipment capable of receiving data from a computing unit and producing converting these data into output controlling one or more hardware devices influencing the

granulation process, such as feed streams, speed, temperature, airflows etc.

The procedure for carrying out the present is demonstrated in the following experiments. The experiments are only examples on embodiments of the invention and should in no way be interpreted as limiting to the scope of the invention.

EXAMPLES

Equipment used in the examples:

Digital recorder	SONY	type PC216A
Digital recorder	HHB	type PDR1000
Accelerometer (1)	Endevco	type 2258A
Accelerometers (5/6)	Brûel & Kjær	type 4502/4503
Data recording system	Difa	type Scadas
Signal treatment software	LMS	Cada-X, TMON

Example 1

Effects changing amount of filler in a granulation process as measured with acoustic emission analysis.

In continuous high shear mixer granulation process the amount of cellulose filled supplied to the granulation process was changed at a given time. The granulator was mounted with an accelerometer for detection of vibrations. The vibrations was recording during the change filler supply and the recorded data were first treated with FFT (Fast Fourier Transformation) to remove noise. Subsequently the data was processed using Principal component analysis and a score plot was generated as depicted in figure 1. From the score plot is can be seen that

that a change in vibration occurs upon changing the amount of filler supplied to the process.

PCA was performed with The Unscrambler®. The PCA algorithm is described in The Unscrambler User Manual, Camo ASA, 1998. Basic chemometric theory e.g. PCA and scoreplots are given in Martens H., Næs, T., Multivariate calibration, 2. ed, Wiley, New York, 1993 and Esbensen, K. et. al., Multivariate Analysis in practice, 3. ed, Camo ASA, Trondheim, 1998.

Example 2

Different placements of accelerometers.
In this example the effect of different placements of accelletometers were tested when chaning the amount of binder to a continuous granulation process. 4 accellerometers were placed close to each other on the same horizontal line on a high shear mixer granulator and this configuration were expected to give only small time delays between the accelerometers. This was not the case. The first accelerometer on the line detected a change in binder as expected while the second, third and fourth accelerometer did not detect the changes at all or detected them very poorly. This shows that the placement of the accelerometers is important when using acoustic emission for detecting changes in granulate characteristics.

PATENT CLAIMS

1. A method for acoustic emission analysis of a granular composition comprising a biologically active compound, said method comprising colliding the granular composition with at least one surface transmitting low frequency vibrations, recording low frequency vibration data in range of 20 Hz to 20 kHz arising from the collision with at least one vibration detector and subjecting the recorded low frequency vibration data to computerized data processing.

2. The method of claim 1, wherein the low frequency vibrations has a frequency between 500 Hz to 5 kHz.

3. The method of claim 1, wherein the detector is an accelerometer.

4. The method of claim 1, wherein the data processing comprises FFT and PCA.

5. The method of claims 1-4, wherein the biologically active compound is in a purified form.

6. The method of claim 1-4, wherein the biologically active compound is selected from bio-catalysts, therapeutic agents, herbicides, pesticides and fungicides.

7. The method of claim 6, wherein the biologically active compound is selected from proteins and peptides.

8. The method of claim 7, wherein the biologically active compound is an enzyme, preferably a selected from hydrolases and oxidoreductases.

9. The method of claim 1, wherein the granular composition further comprises auxiliary granulation agents.

10. The method of claim 9, wherein the auxiliary granulation agents are selected from fiber materials, binders, fillers, liquid agents, enzyme stabilizers, suspension agents, cross linking agents, mediators and/or solvents

11. The method of claim 1, wherein the granules comprises a core wherein the biologically active compound is intimately mixed with auxiliary granulation agents.

12. The method of claim 1, wherein the granules comprise a core particle coated with a layer comprising the biologically active compound and preferably auxiliary granulation agents.

13. The method of claim 1, wherein the granules have an average size between 20-2000 μm, preferably between 100-1000 μm, more preferably between 200-800 μm.

14. The method of claim 1, wherein the granules are coated with a coating agent.

15. A process for preparing granules comprising a biologically active compound and optionally auxiliary granulation agents in a granulation apparatus said process comprising the step of performing acoustic emission analysis on the granules in accordance with claims 1-14 on the granules forming in the granulator.

16. The process of claim 15, wherein the acoustic emission analysis is performed on-line and in real time during the

granulation process and is repeated more than one time during the granulation process.

17. The process of claim 15, further comprising the step of changing at least one process parameter as a result of the acoustic emission analysis.

18. The process of claim 17, wherein process parameter is selected from supply of biologically active compound, supply of auxiliary granulation agent, supply of coating agent, supply of gas, temperature, pressure, pH and mechanical force conferred to the granulation material.

19. The process of claim 15, further characterized by being a coating process wherein granules comprising a biologically active compound and optionally auxiliary granulation agents are coated with a coating agent and the parameter is supply of coating agent to the granulation apparatus.

20. A granulation or coating apparatus comprising

(a) a granulation or coating device comprising at least one chamber for processing material into granules or coated granules,

(b) an arrangement for performing acoustic emission analysis comprising at least one vibration detector capable of detecting vibrations in the range of 20 Hz-20 kHz and optionally amplifying and filtering units and a computer unit.

21. The apparatus of claim 20, wherein the granulating or coating device is selected from a fluid bed granulator or coater, high shear mixer granulator, a coating mixer, a spray dryer, a spray cooler, an extruder.

22. The apparatus of claim 21, further comprising means for providing a purge stream of granules from the chamber and wherein the optical arrangement is positioned to allow fluorescence analysis of granules in the purge stream.

23. The apparatus of claim 21, further comprising one or more elements selected form computing units and control units

24. Use of acoustic emission analysis on granules comprising a biologically active compound.

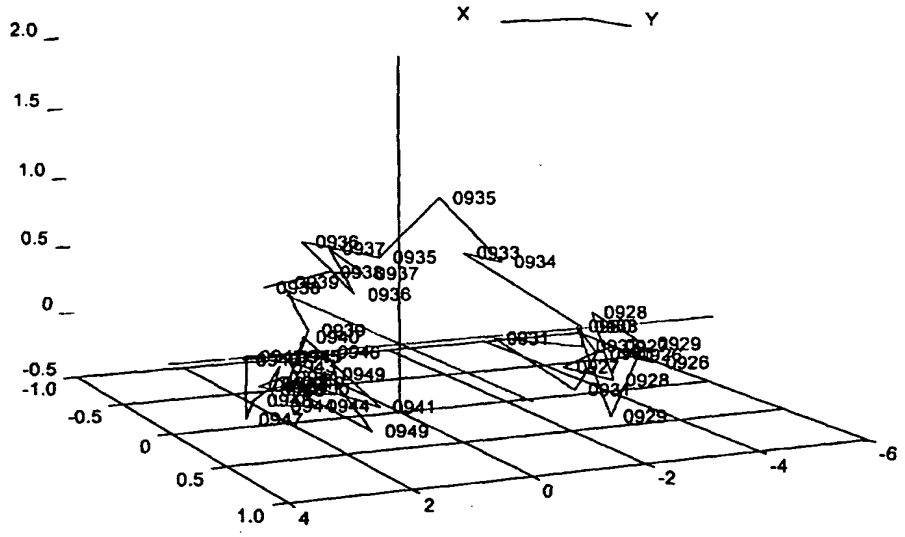
ABSTRACT

The present invention relates to a method for acoustic emission analysis of a granular composition comprising a biologically active compound, said method comprising colliding the granular composition with at least one surface transmitting low frequency vibrations, recording low frequency vibration data in range of 20 Hz to 20 kHz arising from the collision with at least one vibration detector and subjecting the recorded low frequency vibration data to computerized data processing.

DRAWINGS

Figure 1

Scores



B1A 2A, X-expl: 75%,2%,2%